

Supporting information

Genome mining of non-conventional yeasts: search and analysis of *MAL* clusters and proteins

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Table S1. α -glucosidases (AGs) used in current study

Yeast/Bacterium	Acronym for α -glucosidase	Mycocosm ID	GenBank ID	Protein length (aa)	No of introns
<i>Blastobotrys adenivorans</i>	AG1	rna_ARAD1C14212g	Not available	567	0
	AG2	rna_ARAD1D20130g	Not available	581	0
<i>Lipomyces starkeyi</i>	AG1	gm1.3262_g	ODQ72902	582	2
	AG2	fgenes1_kg.8_#_10_#_Locus10580v1rpkm1.26	ODQ71836.1	621	4
	AG3	fgenes1_kg.21_#_1_#_Locus3996v1rpkm38.55	ODQ68842	582	6
	AG4	CE77125_4419	ODQ69382.1	578	4
	AG5	fgenes1_kg.10_#_105_#_Locus4759v1rpkm25.91	ODQ71069.1	641	4
	AG6	fgenes1_kg.6_#_581_#_Locus4531v6rpkm0.18_PRE	ODQ73295.1	574	0
	AG7	estExt_Genemark1.C_170108	ODQ69164.1	629	1
	AG8	gm1.6616_g	ODQ69958.1	555	1
<i>Cyberlindnera fabianii</i>	AG1.1	Not available	CDR39670.1	563	0
	AG1.2	Not available	CDR39674.1	563	0
<i>Meyerozyma guilliermondii</i>	AG1	PGUG_01643T0	EDK37545.2	567	0
	AG2	PGUG_05731T0	XP_001481968.1	568	0
<i>Torulaspora delbrueckii</i>	AG1	TDEL_0D06530	XP_003681448.1	604	0
<i>Saccharomyces cerevisiae</i>	MAL32	YBR299W	NP_009858.3	584	0
	IMA1	YGR287C	NP_011803.3	589	0
<i>Ogataea polymorpha</i>	MAL1	fgenes1_kg.1_#_333_#_isotig01325	XP_018213389.1	564	0
<i>Ogataea parapolyomorpha</i>	AG1	HPODL_02607T0	XP_013932394	564	0
<i>Scheffersomyces stipitis</i>	AGL1	e_gww1.6.1.354.1	XP_001385341.1	572	0
	MAL6	e_gwh1.2.1.484.1	XP_001382912.2	572	0
	MAL7	e_gww1.5.1.399.1	XP_001384657.2	572	0
	MAL8	estExt_genewise1_worm.C_chr_6.10336	XP_001385796.1	572	0
	MAL9	fgenes1_pg.C_chr_1.2000465	XP_001387228.2	573	0
<i>Lodderomyces elongisporus</i>	AG1	LELG_01359T0	XP_001526531.1	585	0
<i>Debaryomyces hansenii</i>	AG1	Not available	XP_459350.2	578	0
<i>Schizosaccharomyces pombe</i>	Mal1	mal1	NP_595063.1	579	0
<i>Bacillus stearothermophilus</i>	α -1,4-glucosidase	Not applicable	BAA12704.1	555	0
<i>Bacillus thermoglucosidasius</i>	oligo-1,6-glucosidase	Not applicable	P29094.1	562	0
<i>Aspergillus oryzae</i>	MalT	AO090038000234_mRNA	XP_001825184.1	574	0
<i>Aspergillus niger</i>	AgdC	An02g13240m.01	XP_001400455.1	587	2
<i>Fusarium oxysporum</i>	Foagl1	FOXG_00152T1	XP_018231823	571	4

Table S2. α -glucoside transporters (AGTs) used in current study

Yeast	Acronym for α -glucoside transporter	Mycocosm ID	GenBank ID	Protein length (aa)	No of introns
<i>Blastobotrys adenivorans</i>	AGT1	rna_ARAD1D09658g	Not available	534	0
<i>Lipomyces starkeyi</i>	AGT1	estExt_Genewise1.C_6_t10076	ODQ72903	510	0
	AGT2.1	gm1.4217_g	ODQ71837.1	511	1
	AGT2.2	estExt_Genewise1.C_8_t10022	ODQ71838.1	498	0
	AGT3	estExt_Genemark1.C_210003	ODQ68844	533	1
	AGT4	fgenesh1_kg.16_#_243_#_Locus11337v1rpkm1.02	ODQ69380.1	616	0
	AGT5	gm1.5097_g	ODQ71068.1	527	1
	AGT6	gm1.3625_g	ODQ73296.1	544	1
<i>Cyberlindnera fabianii</i>	AGT1	Not available	CDR39675.1	582	0
<i>Meyerozyma guilliermondii</i>	AGT1.1	PGUG_01640T0	EDK37542.2	579	0
	AGT1.2	PGUG_01641T0	EDK37543.2	585	0
	AGT1.3	PGUG_01642T0	XP_001485971.1	574	0
<i>Torulaspora delbrueckii</i>	AGT1	TDEL_0D06520	XP_003681447.1	614	0
<i>Saccharomyces cerevisiae</i>	MAL61*	NA	P15685.1	614	0
	AGT1	YGR289C	NP_011805.3	616	0
	MPH2	YDL247W	P0CD99.1	609	0
	MPH3	YJR160C	P0CE00.1	602	0
<i>Ogataea polymorpha</i>	MAL2	fgenesh1_kg.1_#_334_#_isotig00883	XP_018213391.1	522	0
<i>Ogataea parapolyomorpha</i>	AGT1	HPODL_02606T0	XP_013932393	582	0
<i>Scheffersomyces stipitis</i>	MAL1	fgenesh1_pg.C_chr_2.1000315	XP_001382383.1	581	0
	MAL2	fgenesh1_pg.C_chr_5.1000002	XP_001385023.1	585	0
	MAL3	e_gwh1.6.1.324.1	XP_001385693.1	581	0
	MAL4	e_gwh1.6.1.358.1	XP_001385456.1	583	0
	MAL5	fgenesh1_pg.C_chr_6.1000012	XP_001385340.2	509	0
<i>Lodderomyces elongisporus</i>	AGT1	LELG_01361T0	XP_001526533.1	584	0
<i>Debaryomyces hansenii</i>	AGT1	DEHA2E00550g	XP_459351.2	582	0
<i>Schizosaccharomyces pombe</i>	Sut1	sut1	NP_001342860.1	553	1
<i>Aspergillus oryzae</i>	MalP	AO090038000233_mRNA	XP_001825183.1	539	0

* Strain CB11

Table S3. Identity matrix of α - glucosidases (AGs) of yeasts and bacilli																																
	<i>T. delbrueckii</i> AG1	<i>S. cerevisiae</i> MAL32	<i>S. cerevisiae</i> IMA1	<i>C. fabianii</i> AG1.1	<i>C. fabianii</i> AG1.2	<i>O. parapolymorpha</i> AG1	<i>O. polymorpha</i> MAL1	<i>M. guilliermondii</i> AG1	<i>M. guilliermondii</i> AG2	<i>L. elongisporus</i> AG1	<i>S. stipitis</i> AGL1	<i>S. stipitis</i> MAL6	<i>S. stipitis</i> MAL7	<i>S. stipitis</i> MAL8	<i>S. stipitis</i> MAL9	<i>D. hansenii</i> AG1	<i>L. starkeyi</i> AG1	<i>L. starkeyi</i> AG2	<i>L. starkeyi</i> AG3	<i>L. starkeyi</i> AG4	<i>L. starkeyi</i> AG5	<i>L. starkeyi</i> AG6	<i>L. starkeyi</i> AG7	<i>L. starkeyi</i> AG8	<i>B. adenivorans</i> AG1	<i>B. adenivorans</i> AG2	<i>S. pombe</i> Mal1	<i>A. oryzae</i> MalT	<i>A. niger</i> AgdC	<i>F. oxysporum</i> Foag1	<i>Bs</i> α -1,4-glucosidase	<i>Bt</i> oligo-1,6-glucosidase
<i>T. delbrueckii</i> AG1	100	68	81	42	41	41	41	47	49	46	47	47	46	47	46	41	44	34	37	36	34	44	34	35	38	41	37	43	37	35	35	40
<i>S. cerevisiae</i> MAL32	68	100	72	42	44	43	43	47	49	46	49	49	47	48	41	44	34	36	36	33	44	34	36	40	41	38	43	36	35	37	39	
<i>S. cerevisiae</i> IMA1	81	72	100	44	43	44	44	49	50	48	49	51	49	50	44	45	34	38	37	34	46	35	35	40	44	38	44	37	36	36	42	
<i>C. fabianii</i> AG1.1	42	42	44	100	77	76	75	56	57	54	56	57	58	56	41	46	38	39	38	37	44	36	38	41	44	40	47	39	39	40	42	
<i>C. fabianii</i> AG1.2	41	44	43	77	100	86	86	56	58	57	59	58	59	56	43	46	38	38	38	37	43	38	39	40	46	40	47	39	40	41	42	
<i>O. parapolymorpha</i> AG1	41	43	44	76	86	100	98	57	59	56	59	60	60	59	57	41	46	38	38	37	36	43	36	38	39	44	40	45	38	38	40	41
<i>O. polymorpha</i> MAL1	41	43	44	75	86	98	100	57	59	56	58	59	59	56	41	45	38	38	38	37	43	37	37	38	44	40	45	38	38	40	42	
<i>M. guilliermondii</i> AG1	47	47	49	56	56	57	57	100	62	59	63	64	62	63	60	45	51	39	41	41	39	46	39	39	42	48	43	51	43	42	39	44
<i>M. guilliermondii</i> AG2	49	49	50	57	58	59	59	62	100	66	65	67	66	66	71	45	53	40	42	43	40	49	41	42	43	49	45	51	43	43	39	45
<i>L. elongisporus</i> AG1	46	46	48	54	57	56	56	59	66	100	65	69	68	68	63	43	49	38	38	39	37	47	37	38	39	47	42	50	40	42	37	42
<i>S. stipitis</i> AGL1	47	49	49	56	59	59	58	63	65	65	100	79	76	77	66	42	51	37	40	40	36	46	37	39	40	45	42	52	40	42	41	43
<i>S. stipitis</i> MAL6	47	49	51	57	58	60	59	64	67	69	79	100	85	83	68	43	52	39	40	40	39	47	38	39	42	44	44	53	41	43	39	44
<i>S. stipitis</i> MAL7	46	49	49	58	58	60	59	62	66	68	76	85	100	79	68	43	51	38	40	41	37	47	38	39	40	45	42	53	41	43	40	44
<i>S. stipitis</i> MAL8	47	47	50	56	59	59	59	63	66	68	77	83	79	100	66	44	50	38	40	41	39	47	38	39	41	45	44	52	42	43	39	43
<i>S. stipitis</i> MAL9	46	48	49	57	56	57	56	60	71	63	66	68	68	66	100	43	50	38	42	43	38	48	38	40	41	48	43	51	41	42	40	43
<i>D. hansenii</i> AG1	41	41	44	41	43	41	41	45	45	43	42	43	43	44	43	100	45	51	56	59	52	43	52	58	49	45	51	43	54	53	41	46
<i>L. starkeyi</i> AG1	44	44	45	46	46	46	45	51	53	49	51	52	51	50	50	45	100	42	43	47	41	54	41	43	43	50	45	55	45	44	40	43
<i>L. starkeyi</i> AG2	34	34	34	38	38	38	38	39	40	38	37	39	38	38	38	51	42	100	53	59	74	39	56	58	42	40	45	42	46	46	36	42
<i>L. starkeyi</i> AG3	37	36	38	39	38	38	38	41	42	38	40	40	40	40	42	56	43	53	100	58	53	43	53	58	49	44	49	44	56	55	39	47
<i>L. starkeyi</i> AG4	36	36	37	38	38	37	38	41	43	39	40	40	41	41	43	59	47	59	58	100	59	43	58	64	50	44	51	45	55	52	40	48
<i>L. starkeyi</i> AG5	34	33	34	37	37	36	37	39	40	37	36	39	37	39	38	52	41	74	53	59	100	39	57	58	44	40	45	40	48	46	36	42
<i>L. starkeyi</i> AG6	44	44	46	44	43	43	43	46	49	47	46	47	47	47	48	43	54	39	43	43	39	100	41	41	44	49	41	56	44	42	39	41
<i>L. starkeyi</i> AG7	34	34	35	36	38	36	37	39	41	37	37	38	38	38	38	52	41	56	53	58	57	41	100	69	45	41	45	43	49	49	37	42
<i>L. starkeyi</i> AG8	35	36	35	38	39	38	37	39	42	38	39	39	39	39	40	58	43	58	58	64	58	41	69	100	49	42	49	44	52	53	40	45
<i>B. adenivorans</i> AG1	38	40	40	41	40	39	38	42	43	39	40	42	40	41	41	49	43	42	49	50	44	44	45	49	100	46	57	44	49	47	39	44
<i>B. adenivorans</i> AG2	41	41	44	44	46	44	44	48	49	47	45	44	45	45	48	45	50	40	44	44	40	49	41	42	46	100	41	49	44	41	38	40
<i>S. pombe</i> Mal1	37	38	38	40	40	40	40	43	45	42	42	44	42	44	43	51	45	45	49	51	45	41	45	49	57	41	100	42	50	49	39	42
<i>A. oryzae</i> MalT	43	43	44	47	47	45	45	51	51	50	52	53	53	52	51	43	55	42	44	45	40	56	43	44	44	49	42	100	44	44	41	42
<i>A. niger</i> AgdC	37	36	37	39	39	38	38	43	43	40	40	41	41	42	41	54	45	46	56	55	48	44	49	52	49	44	50	44	100	57	39	45
<i>F. oxysporum</i> Foag1	35	35	36	39	40	38	38	42	43	42	42	43	43	43	42	53	44	46	55	52	46	42	49	53	47	41	49	44	57	100	40	48
<i>Bs</i> α -1,4-glucosidase	35	37	36	40	41	40	40	39	39	37	41	39	40	39	40	41	40	36	39	40	36	39	37	40	39	38	39	41	39	40	100	56
<i>Bt</i> oligo-1,6-glucosidase	40	39	42	42	42	41	42	44	45	42	43	44	44	43	43	46	43	42	47	48	42	41	42	45	44	40	42	42	45	48	56	100

Bs, *Bacillus stearothermophilus*; *Bt*, *Bacillus thermoglucosidasius*

Table S4. Identity matrix of yeast α-glucoside transporters (AGTs)																													
	<i>T. delbrueckii</i> AGT1	<i>S. cerevisiae</i> MAL61	<i>S. cerevisiae</i> MPH2	<i>S. cerevisiae</i> MPH3	<i>S. cerevisiae</i> AGT1	<i>C. fabianii</i> AGT1	<i>O. parapolymorpha</i> AGT1	<i>O. polymorpha</i> MAL2	<i>M. guilliermondii</i> AGT1.1	<i>M. guilliermondii</i> AGT1.2	<i>M. guilliermondii</i> AGT1.3	<i>L. elongisporus</i> AGT1	<i>D. hansenii</i> AGT1	<i>S. stipitis</i> MAL1	<i>S. stipitis</i> MAL2	<i>S. stipitis</i> MAL3	<i>S. stipitis</i> MAL4	<i>S. stipitis</i> MAL5	<i>L. starkeyi</i> AGT1	<i>L. starkeyi</i> AGT2.1	<i>L. starkeyi</i> AGT2.2	<i>L. starkeyi</i> AGT3	<i>L. starkeyi</i> AGT4	<i>L. starkeyi</i> AGT5	<i>L. starkeyi</i> AGT6	<i>A. oryzae</i> MalP	<i>B. adenivorans</i> AGT1	<i>S. pombe</i> Sut1	<i>S. pombe</i> Ght3
<i>T. delbrueckii</i> AGT1	100	53	52	52	67	36	36	35	39	37	37	36	36	36	36	37	35	26	32	14	29	33	34	18	34	33	32	14	9
<i>S. cerevisiae</i> MAL61	53	100	73	74	54	36	38	37	42	40	39	38	37	38	39	38	38	27	36	17	29	32	35	19	36	36	32	16	8
<i>S. cerevisiae</i> MPH2	52	73	100	96	51	36	39	37	40	40	38	39	38	37	38	38	36	27	37	16	29	33	36	19	36	37	31	16	7
<i>S. cerevisiae</i> MPH3	52	74	96	100	52	36	39	38	41	40	38	39	38	37	38	37	36	28	37	16	30	33	36	19	37	37	31	16	7
<i>S. cerevisiae</i> AGT1	67	54	51	52	100	38	37	37	39	39	35	37	37	37	38	37	37	28	35	15	28	33	36	20	34	35	31	16	8
<i>C. fabianii</i> AGT1	36	36	36	36	38	100	73	67	57	55	54	51	49	55	51	52	54	30	41	17	31	41	42	20	41	41	38	16	9
<i>O. parapolymorpha</i> AGT1	36	38	39	39	37	73	100	87	57	56	54	50	50	52	51	51	51	29	41	18	31	39	40	20	41	41	37	17	9
<i>O. polymorpha</i> MAL2	35	37	37	38	37	67	87	100	54	53	53	48	47	50	49	49	49	33	45	19	35	40	41	20	41	42	38	18	9
<i>M. guilliermondii</i> AGT1.1	39	42	40	41	39	57	57	54	100	79	69	55	56	58	56	54	55	30	43	18	34	40	43	22	42	42	40	17	10
<i>M. guilliermondii</i> AGT1.2	37	40	40	40	39	55	56	53	79	100	68	56	55	55	55	52	56	31	41	18	34	39	42	22	42	41	38	17	9
<i>M. guilliermondii</i> AGT1.3	37	39	38	38	35	54	54	53	69	68	100	53	54	55	53	53	53	31	39	19	32	40	42	22	41	41	38	17	10
<i>L. elongisporus</i> AGT1	36	38	39	39	37	51	50	48	55	56	53	100	50	63	61	61	63	29	41	18	32	39	40	19	42	42	38	15	8
<i>D. hansenii</i> AGT1	36	37	38	38	37	49	50	47	56	55	54	50	100	48	48	49	49	30	39	18	31	39	40	20	40	39	35	16	9
<i>S. stipitis</i> MAL1	36	38	37	37	37	55	52	50	58	55	55	63	48	100	88	81	83	31	41	18	33	39	40	20	41	42	38	17	8
<i>S. stipitis</i> MAL2	36	39	38	38	38	51	51	49	56	55	53	61	48	88	100	77	79	30	41	18	32	39	39	19	42	41	37	17	8
<i>S. stipitis</i> MAL3	37	38	38	37	37	52	51	49	54	52	53	61	49	81	77	100	76	30	42	17	33	40	39	20	41	42	37	17	8
<i>S. stipitis</i> MAL4	35	38	36	36	37	54	51	49	55	56	53	63	49	83	79	76	100	30	39	18	31	38	37	20	40	42	37	16	8
<i>S. stipitis</i> MAL5	26	27	27	28	28	30	29	33	30	31	31	29	30	31	30	30	30	100	37	19	45	32	30	19	34	35	35	17	7
<i>L. starkeyi</i> AGT1	32	36	37	37	35	41	41	45	43	41	39	41	39	41	41	42	39	37	100	19	38	45	43	20	47	47	41	17	8
<i>L. starkeyi</i> AGT2.1	14	17	16	16	15	17	18	19	18	18	19	18	18	18	17	18	19	19	19	100	19	17	16	19	17	19	18	19	7
<i>L. starkeyi</i> AGT2.2	29	29	29	30	28	31	31	35	34	34	32	32	31	33	32	33	31	45	38	19	100	34	33	23	36	35	38	18	10
<i>L. starkeyi</i> AGT3	33	32	33	33	33	41	39	40	40	39	40	39	39	39	39	40	38	32	45	17	34	100	65	18	47	47	40	16	8
<i>L. starkeyi</i> AGT4	34	35	36	36	36	42	40	41	43	42	42	40	40	40	39	39	37	30	43	16	33	65	100	18	43	44	38	15	7
<i>L. starkeyi</i> AGT5	18	19	19	19	20	20	20	20	22	22	22	19	20	20	19	20	20	19	20	19	23	18	18	100	22	22	22	18	7
<i>L. starkeyi</i> AGT6	34	36	36	37	34	41	41	41	42	42	41	42	40	41	42	41	40	34	47	17	36	47	43	22	100	51	44	17	9
<i>A. oryzae</i> MalP	33	36	37	37	35	41	41	42	42	41	41	42	39	42	41	42	42	35	47	19	35	47	44	22	51	100	43	16	10
<i>B. adenivorans</i> AGT1	32	32	31	31	31	38	37	38	40	38	38	38	35	38	37	37	37	35	41	18	38	40	38	22	44	43	100	17	8
<i>S. pombe</i> Sut1	14	16	16	16	16	16	17	18	17	17	17	15	16	17	17	17	16	17	17	19	18	16	15	18	17	16	17	100	4
<i>S. pombe</i> Ght3	9	8	7	7	8	9	9	9	10	9	10	8	9	8	8	8	8	7	8	7	10	8	7	7	9	10	8	4	100

Table S5. Primers for amplification of *Scheffersomyces stipitis* α -glucosidase genes of the *MAL* loci

Gene	5'-3'	
	Forward	Reverse
MAL7	ACCACTCTAGAAAGTAATAAATTCC	GAAAATTATCTTCGATTCATATTTAAGG
MAL8	GATCTTCATGTATTCATAAATATTTGTACG	CCAATCCTTCTATGTATCTTCC
MAL9	ATCTCATATTTTCAGAATCAACTACTC	GGTACTCTGGAGGACCCG

Table S6. Primers for cloning the *Scheffersomyces stipitis* α -glucosidase genes to pURI3-Cter vector [1]

Gene	5'-3'	
	Forward	Reverse
MAL7	TAACTTTAAGAAGGAGATATACATATGACAATTGCTCGAGAATGGTGG	GCTATTAATGATGATGATGATGATGATCAACAATGTATAGACGACCTT
MAL8	TAACTTTAAGAAGGAGATATACATATGACTGTTGCACACAAATGGTGG	TAACTTTAAGAAGGAGATATACATATGACTGTTGCACACAAATGGTGG
MAL9	TAACTTTAAGAAGGAGATATACATATGACTAAAAGAATCTGGTGGAAAG	GCTATTAATGATGATGATGATGATGGGACAACAAGTAGACCCGGGCT

Table S7. Synteny between the *Ogataea polymorpha* NCYC 495 and *Ogataea parapolymorpha* DL-1 genomes according to MycoCosm data. Forward synteny between the chromosomes is shown on white background, reverse synteny on grey background.

Yeast	Chromosome No (length in Mbp)						
<i>Ogataea polymorpha</i> NCYC 495 leu1.1	1 (1.54)	2 (1.52)	3 (1.36)	4 (1.30)	5 (1.27)	6 (1.00)	7 (0.99)
<i>Ogataea parapolymorpha</i> DL-1*	7 (1.51)	6 (1.51)	5 (1.33)	4 (1.29)	3 (1.27)	2 (0.99)	1 (1.14)

* Chromosome lengths were taken from [2]

Table S8. Hypothetical permease and α -glucosidase genes outside the *MAL* locus in *Ogataea polymorpha* and *Ogataea parapolymorpha* with their genomic neighbors indicated. Transcription direction is indicated by arrow.

<i>Ogataea</i> strains (Chromosomes)	Genes and potential function of proteins		
	copper transporter	α -glucosidase	gamma-tubulin
<i>O. polymorpha</i> NCYC 495 leu1.1 (Chr 7)	→	←	→
<i>O. parapolymorpha</i> DL-1 (Chr 1)	→	←	→
<i>Ogataea</i> strains (Chromosomes)	Genes and potential function of proteins		
	clathrin coat assembly protein	α -glucoside permease	aminopeptidase
<i>O. polymorpha</i> NCYC 495 leu1.1 (Chr 6)	←	→	←
<i>O. parapolymorpha</i> DL-1 (Chr 2)	←	→	←

References

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2. Ravin, N. V.; Eldarov, M. A.; Kadnikov, V. V.; Beletsky, A. V.; Schneider, J.; Mardanova, E. S.; Smekalova, E. M.; Zvereva, M. I.; Dontsova, O. A.; Mardanov, A. V.; Skryabin, K. G. Genome sequence and analysis of methylotrophic yeast *Hansenula polymorpha* DL1. *BMC Genomics* **2013**, *14*, 837.